PCT

WORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 6 :
C07K 14/47, C12N 15/12, A61K 38/17, C12N 15/10, C12P 21/02, A61K 48/00

A2

(11) International Publication Number: WO 99/32516

(43) International Publication Date: 1 July 1999 (01.07.99)

(21) International Application Number: PCT/IT98/00364

(22) International Filing Date: 16 December 1998 (16.12.98)

(30) Priority Data:
RM97A000796 19 December 1997 (19.12.97) IT

(71) Applicant *(for all designated States except US)*: SIGMA-TAU INDUSTRIE FARMACEUTICHE RIUNITE S.P.A. [IT/IT]; Viale Shakespeare, 47, I-00144 Rome (IT).

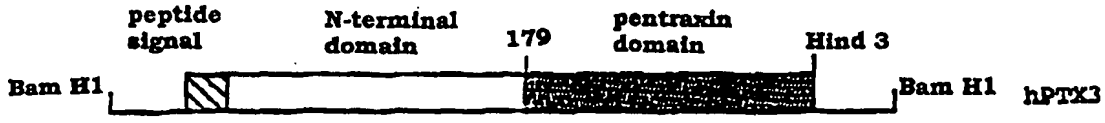
(72) Inventors; and
(75) Inventors/Applicants *(for US only)*: BOTTAZZI, Barbara [IT/IT]; Via Cascina Bianca, 28/2, I-20142 Milan (IT). INTRONA, Martino [IT/IT]; Via Balzarotti, 12/I, I-20017 Rho (IT). MANTOVANI, Alberto [IT/IT]; Largo Brasilia, 4, I-20100 Milan (IT). VECCHI, Annunciata [IT/IT]; Via Spontini, 11, I-20131 Milan (IT).

(74) Common Representative: SIGMA-TAU INDUSTRIE FARMACEUTICHE RIUNITE S.P.A.; Viale Skakespeare, 47, I-00144 Rome (IT).

(81) Designated States: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, HR, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published
Without international search report and to be republished upon receipt of that report.

(54) Title: PHARMACEUTICAL COMPOSITIONS CONTAINING THE LONG PENTRAXIN PTX3



(57) Abstract

Pharmaceutical compositions are described containing a long pentraxin PTX3, particularly human PTX3, for the therapy of infectious and inflammatory or tumour diseases; expression vectors containing cDNA coding for PTX3; recombinant host cells transfected with such vectors; a method for producing substantial amounts of PTX3 involving the culturing of such cells, and the use of said vectors in the gene therapy of tumours.

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania
AM	Armenia
AT	Austria
AU	Australia
AZ	Azerbaijan
BA	Bosnia and Herzegovina
BB	Barbados
BE	Belgium
BF	Burkina Faso
BG	Bulgaria
BJ	Benin
BR	Brazil
BY	Belarus
CA	Canada
CF	Central African Republic
CG	Congo
CH	Switzerland
CI	Côte d'Ivoire
CM	Cameroon
CN	China
CU	Cuba
CZ	Czech Republic
DE	Germany
DK	Denmark
EE	Estonia
ES	Spain
FI	Finland
FR	France
GA	Gabon
GB	United Kingdom
GE	Georgia
GH	Ghana
GN	Guinea
GR	Greece
HU	Hungary
IE	Ireland
IL	Israel
IS	Iceland
IT	Italy
JP	Japan
KE	Kenya
KG	Kyrgyzstan
KP	Democratic People's Republic of Korea
KR	Republic of Korea
KZ	Kazakstan
LC	Saint Lucia
LI	Liechtenstein
LK	Sri Lanka
LR	Liberia
LS	Lesotho
LT	Lithuania
LU	Luxembourg
LV	Latvia
MC	Monaco
MD	Republic of Moldova
MG	Madagascar
MK	The former Yugoslav Republic of Macedonia
ML	Mali
MN	Mongolia
MR	Mauritania
MW	Malawi
MX	Mexico
NE	Niger
NL	Netherlands
NO	Norway
NZ	New Zealand
PL	Poland
PT	Portugal
RO	Romania
RU	Russian Federation
SD	Sudan
SE	Sweden
SG	Singapore
SI	Slovenia
SK	Slovakia
SN	Senegal
SZ	Swaziland
TD	Chad
TG	Togo
TJ	Tajikistan
TM	Turkmenistan
TR	Turkey
TT	Trinidad and Tobago
UA	Ukraine
UG	Uganda
US	United States of America
UZ	Uzbekistan
VN	Viet Nam
YU	Yugoslavia
ZW	Zimbabwe

Pharmaceutical compositions containing the long pentraxin PTX3

The present invention relates to pharmaceutical compositions containing the long pentraxin PTX3 (PTX3) or one of its functional derivatives. In particular, the invention relates to the aforesaid compositions for the therapy of infectious and inflammatory diseases or tumours.

The invention also relates to expression vectors containing the complete cDNA sequence coding for PTX3 or one of its functional derivatives, recombinant host cells transfected with such expression vectors and a method for producing PTX3 or one of its functional derivatives. Further, the invention relates to gene therapy methods for the treatment of tumours, based on the use of the aforesaid expression vectors.

To date, we have yet to fully understand the biological function of PTX3, a protein which is expressed in various types of cells, most notably in mononuclear phagocytes and endothelial cells, after exposure to the inflammatory cytokines Interleukin 1beta (IL-1beta) and Tumour Necrosis Factor alpha (TNF-alpha).

To date, there has also been no description of any therapeutic use of PTX3 or of its functional derivatives.

PTX3 consists of two structural domains, an N-terminal unrelated to any known molecule, and a C-terminal similar to the short pentraxins such as C-reactive protein (CRP). A substantial degree of similarity has been found between human PTX3 (hPTX3) and animal PTX3s.

The PTX3 gene is located on chromosome 3 of the mouse in a region similar to the human 3q region (q24-28), in agreement with the documented location of hPTX3 in the 3q 25 region. Furthermore, mouse PTX3 (mPTX3) (Introna M., Vidal Alles V., Castellano M., Picardi G., De Gioia L., Bottazzi B., Peri G., Breviario F., Salmona M., De Gregorio L., Dragani T.A., Srinivasan N., Blundell T.L., Hamilton T.A. and Mantovani A.: Cloning of mouse PTX3, a new member of the pentraxin gene family expressed at extrahepatic sites. Blood 87 (1996) 1862-1872) is very similar to hPTX3 in terms of organisation, location and sequence (Breviario F., d'Aniello E.M., Golay J., Peri G., Bottazzi B., Bairoch A., Saccone S., Marzella R., Predazzi V., Rocchi M., Della Valle G., Dejana E., Mantovani A., Introna M.: Interleukin-1-inducible genes in endothelial cells. Cloning of a new gene related to C-reactive protein and serum amyloid P component. J. Biol. Chem. 267:22190, 1992).

In particular, the degree of identity between the sequences is 82% between the human and mouse gene and reaches 90% if conservative substitutions are considered.

The high degree of similarity between the hPTX3 and mPTX3 sequences is a sign of the high degree of conservation of pentraxin during evolution (Pepys M.B., Baltz M.L.: Acute phase proteins with special reference to C-reactive protein and related proteins (pentraxins) and serum amyloid A protein. Adv. Immunol. 34:141, 1983).

CRP is a marker for immuno-inflammatory and infectious disease. After a trauma, a lesion or infection of a tissue triggers off, in the affected subject, a complex series of reactions aimed at preventing extension of the damage, at destroying the infecting organism and at activating the repair process in order to restore normal function. This process constitutes the so-called acute-phase response, and the main marker currently used for the acute-phase response is CRP. In normal human serum, in fact, it is present in concentrations of less than 10 μg/ml, but can increase more than 1,000-fold in response to a trauma or inflammation (Koj A.: "Acute phase reactants" in "Structure and Function of Plasma Proteins". Allison A., ed. Plenum Press, New York, 1974, pp. 73-131).

Previous therapeutic uses of CRP are already known. For instance, US Patent 4,857,314 dated 15.08.1989 discloses the use of CRP in combination with TNF for the treatment of tumours.

International patent application PCT/US94/02181 dated 24.02.1994 discloses mutants of CRP which are useful for the preparation of diagnostic kits for determining immunocomplexes in

biological fluids and for the treatment of viral and microbial diseases, tumours and endotoxic shock.

International patent application PCT/US94/09729 dated 26.08.1994 also discloses mutants of CRP which are useful for the preparation of diagnostic kits and for the treatment of viral and microbial diseases and tumours.

The ability of PTX3 to recognise and bind specifically to ligands which are also recognised by short pentraxins has been evaluated *in vitro* using purified recombinant PTX3. Short pentraxins such as CRP and SAP (serum amyloid P component) are characterised by their ability to recognise and bind in a calcium-dependent manner to a broad spectrum of ligands, including phosphocholine, phosphoethanolamine, many sugars, the best characterised of which is an agarose derivative rich in pyruvate [methyl 4-6-O-(1-carboxyethylidene)-beta-D-galacto-pyranoside] or MOβDG, complement fragments and proteins of the extracellular matrix, particularly fibronectin and type IV collagen. Unlike the short pentraxins, PTX3 is unable to bind either calcium (assessed by Inductive Coupled Plasma/Atomic Emission Spectroscopy) or phosphocholine, phosphoethanolamine or MOβDG. Moreover, PTX3 is unable to bind extracellular matrix proteins such as fibronectin or type IV collagen. On the other hand, PTX3 is capable of binding the C1q complement fragment which is also recognised by the short pentraxins (Table 1). It

should be stressed, however, that, whereas CRP and SAP have to be cross-linked to bind C1q, PTX3 is capable of recognising and binding this complement fragment in the naturally occurring form.

Surprisingly, it has now been found that the long pentraxin PTX3 or its functional derivatives are useful therapeutic agents, particularly for the therapy of infectious and inflammatory diseases or tumours.

What is meant by "long pentraxin PTX3" is any long pentraxin PTX3, i.e. regardless of its natural (human or animal) or synthetic origin. Human long pentraxin PTX3 (see sequence 1 and Fig. 5) is the preferred form.

A convenient method of producing substantial amounts of long pentraxin PTX3 or one of its functional derivatives consists in preparing expression vectors (e.g. plasmids) containing the complete cDNA sequence coding for PTX3 or one of its functional derivatives and in using these to transfer eukaryotic cells in culture (e.g. Chinese hamster ovary cells, CHO). After cloning the recombinant host cells thus transfected, the cell clone capable of producing the highest levels of PTX3 is selected.

According to the present invention, the above-mentioned expression vectors containing the cDNA sequence coding for long pentraxin PTX3 are also utilised in gene therapy methods for the treatment of tumour conditions.

A first gene therapy method consists in:

a) taking samples of cells from a patient suffering from a tumour;

b) transfecting these cells with an expression vector containing the complete cDNA sequence coding for long pentraxin PTX3 or one of its functional derivatives; and

c) inoculating the tumour patient with these transfected cells.

A second gene therapy method for the treatment of tumours consists in:

a) preparing an expression vector of viral origin (such as an adenovirus or retrovirus) containing the complete cDNA sequence coding for long pentraxin PTX3 or one of its functional derivatives; and

b) injecting the tumour affected patient with the expression vector thus obtained.

Though the mechanism of action of PTX3 or its functional derivatives has yet to be definitively clarified, their anticancer activity in any event is not attributable to a direct cytolytic or cytostatic effect on the tumour cells, but rather to mechanisms mediated by the host and related to the leukocyte recruitment ability exerted by these compounds, as will be described below.

There now follows a description of the experimental procedures and results are reported demonstrating the unexpected activity of the compounds according to the invention described herein.

Production of recombinant PTX3: a fragment containing the complete cDNA sequence of human PTX3 (sequence 2 and Fig. 6) was subcloned in the Bam H1 site of the expression vector pSG5 (Fig. 1) (Stratagene, La Jolla, CA, USA) and transfected in CHO cells using the precipitated calcium procedure. A clone selected in G418, capable of producing large amounts of PTX3, was used as a source from which the protein was purified by chromatography by means of ion exchange and gel filtration.

Binding of PTX3 to C1q: the binding of PTX3 to C1q was assessed in an ELISA system. A 96-well plate was covered with 250-500 ng of C1q per well (one night at 4°C) and then washed with PBS with Ca^{++} and Mg^{++} containing 0.05% Tween 20 (PBS). The wells were then blocked with 5% milk in PBS (2 h at room temperature) and subsequently incubated with variable concentrations of PTX3 (30 min at 37°C). After a further series of washings, the plate was incubated with a rat monoclonal antibody to PTX3 (1 h at room temperature) and then with the second antibody, a peroxidase-conjugated rat anti-IgG antibody (1 h at room temperature). After washing, chromogen was added and absorbance was read at 405 nm using an automatic plate

reader. In a number of experiments, the wells were covered with PTX3 and C1q binding was evaluated using an anti-C1q antibody.

Biotinylated protein was used to determine the C1q binding affinity. PTX3 was biotinylated according to standard procedures using an activated biotin modified by the addition of a "spacer arm". (SPA – Società Prodotti Antibiotici).

Figures 2(A) and 2(B) give the C1q binding and binding affinity results. These results show the very substantial degree of C1q binding and binding affinity of PTX3.

Leukocyte recruitment: the leukocyte recruitment induced by PTX3 was studied *in vivo* in the subcutaneous pocket system. The subcutaneous pocket was induced in the experimental animal by means of two subcutaneous injections of 5 mL of air into the animal's back with an intervening interval of three days. On day 6, 1 μg of PTX3 in 0.5% carboxymethylcellulose was administered into the pocket. After 4 h, the animals were anaesthetised and the pocket was washed with 1 mL of saline solution. The washing liquid was recovered and was submitted to a total count and a differential count of the cells present.

The results obtained are reported in Figure 3 and show the substantial leukocyte recruitment capacity of PTX3 in normal animals, whereas Figure 4 shows the results obtained in genetically modified

animals, without C1q, in which the leukocyte recruitment is significantly lower.

Anticancer activity: a line of murine mastocytoma P815 was co-transfected by electroporation with the expression vector pSG5 containing the cDNA of human PTX3 or its antisense and the vector pSV2 which endows the transfected cells with neomycin resistance. After selection with neomycin 0.5 mg/mL, the cells were cloned by limit dilution.

To assess the production of PTX3, $2.5x10^5$ cells were cultivated in 200 μL of RPMI + 3% FCS for 24 h and the supernatant was tested by ELISA. The clones obtained produced protein levels ranging from 1 to 35 ng/mL, while the clones containing the antisense produced no measurable levels of PTX3. The clones considered showed the same growth rate *in vivo*.

Male DBA/2N CrlBR mice aged 8-10 weeks were subcutaneously injected with $1x10^5$ cells of P815 PTX3-producing clones or with clones containing the antisense gene. The mice were monitored 3 times daily for occurrence of tumours and once daily for survival.

The results obtained are reported in Table 2 and show the efficacy of PTX3, in this experimental model of gene therapy, in bringing about healing of the animals and complete rejection of the tumour after the take of the inoculated tumour cells.

These results are statistically significant with $p<0.01$ (Fisher test) both as compared to controls and to the group treated with the antisense.

In the light of these results it is clear that the anticancer activity reported above correlates closely with the leukocyte recruitment which occurs in the mouse as a result of recognition of the C1q by PTX3. In genetically modified mice, no such leukocyte recruitment occurs. The leukocyte recruitment capacity, on the basis of the anticancer activity of the compounds according to the invention, indicates that these compounds may also have a useful application in the treatment of diseases caused by pathogens such as bacteria, fungi, protozoa or viruses.

TABLE 1 PENTRAXIN BINDING ABILITY TO VARIOUS LIGANDS

	CRP	SAP	PTX3
Ca^{2+}	+	+	-
Phosphocholine	+	-	-
Phosphoethanolamine	+	+	-
MOβDG	-	+	-
C1q	+	+	+
Type IV collagen	ND	+	-
Fibronectin	ND	+	-

ND: test not performed

TABLE 2 *IN VIVO* ANTICANCER ACTIVITY OF PTX3

Clone[1]	Reject[2]
Parent P815 (control)	4/25
P815-AS1 (antisense)	3/8
P815-PTX3-1 (sense)	14/14*

1 : 1 x 10^5 cells of the clone indicated were injected subcutaneously.

2 : Number of animals that definitely reject the tumour out of total number of animals in which it took.

* : $p<0.01$ as compared both to mice treated with parent cells and to mice treated with cells of the antisense clones (Fisher test).

Brief description of drawings

Figure 2: PTX3 binding to C1q. Panel A shows the binding of the supernatant of the culture containing PTX3 (sense) and of the purified protein to C1q and C1s immobilised on plate. The binding is assessed as optical density (O.D.) at 405 nm. Panel B shows the saturation curve obtained with the biotinylated protein. The kinetic parameters were calculated using the non-linear fitting statistical method.

Figure 3: PTX3-induced leukocyte recruitment: 1 μg of PTX3 is injected into a subcutaneous pocket induced in the back of CD1 mice by inoculation of 5 ml of air.

Figure 4: PTX3-induced leukocyte recruitment in normal animals and in genetically modified animals without C1q. PTX3 is injected into a subcutaneous induced on the back of the animals.

Sequence 1: Amino acid sequence of human PTX3. The underlined amino acids constitute the peptide signal. Mature hPTX3 consists of 364 amino acids.

Sequence 2: Nucleotide sequence of fragment of cDNA of human PTX3. Upper case letters denote nucleotides coding for the protein, while lower case letters denote regions at 3' and 5' not translated but present in the construct.

CLAIMS

1. Orally, parenterally, transdermally or subcutaneously administrable pharmaceutical composition containing as active ingredient the amino acid sequence of the long pentraxin PTX3, and a pharmacologically acceptable excipient.
2. Composition according to claim 1, in which the sequence of the long pentraxin PTX3 is the sequence of naturally occurring PTX3.
3. Composition according to claim 2, in which the sequence of the long pentraxin PTX3 is the sequence of human PTX3.
4. Composition according to claims 1-3, for the treatment of infectious and inflammatory diseases or tumours.
5. Composition according to claim 4, for the treatment of diseases caused by bacteria, fungi, protozoa o viruses.
6. Expression vector containing the complete cDNA sequence coding for the long pentraxin PTX3 or one of its functional derivatives.
7. Vector according to claim 6, which is a plasmid.
8. Vector according to claim 7, which is the plasmid pSG5.
9. Recombinant host cells transfected with the expression vector of claims 6-8.
10. Host cells according to claim 9, which are CHO cells.
11. Method for producing long pentraxin PXT3 or one of its functional derivatives comprising the culturing of the cells of claims 9-10.

12. Method of gene therapy for the treatment of tumour conditions, comprising:

(a) taking samples of cells from a patient suffering from a cancer;

(b) transfecting such cells with an expression vector containing the complete cDNA sequence coding for the long pentraxin PTX3 or one of its functional derivatives; and

(c) inoculating said patient with such transfected cells.

13. Method of gene therapy for the treatment of tumour conditions comprising:

a) preparation of an expression vector of viral origin containing the complete cDNA sequence coding for the long pentraxin PXT3 or one of its functional derivatives; and

b) injection of the expression vector thus obtained into a patient suffering from a cancer.

14. Method according to claim 13, in which the expression vector of viral origin is an adenovirus or retrovirus.

15. Use of the long pentraxin PTX3 for the preparation of a medicament for the treatment of infectious, inflammatory or tumour diseases.

16. Use according to claim 15, in which the long pentraxin PTX3 is the human pentraxin having sequence 1.

17. Use according to claim 16, for the preparation of a medicament for the treatment of diseases caused by bacteria, fungi, protozoa or viruses.

18. Use of cDNA coding for the long pentraxin PTX3 or one of its functional derivatives for the preparation of expression vectors containing such cDNA to be used in gene therapy methods for the treatment of tumour conditions.

FIGURE 1

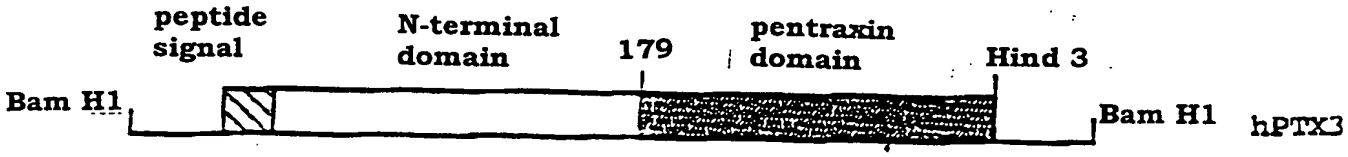
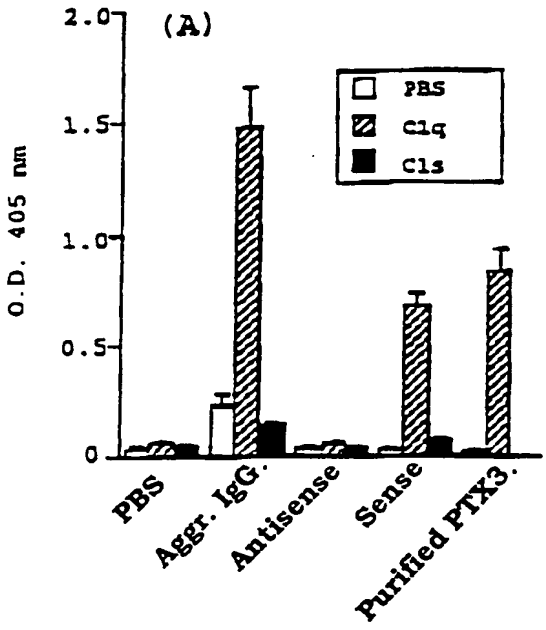

peptide signal
N-terminal domain
179
pentraxin domain
Hind 3
Bam H1
Bam H1
hPTX3

FIGURE 2


(A)
O.D. 405 nm
2.0
1.5
1.0
0.5
0
PBS
C1q
C1s
PBS
Aggr. IgG.
Antisense
Sense
Purified PTX3.

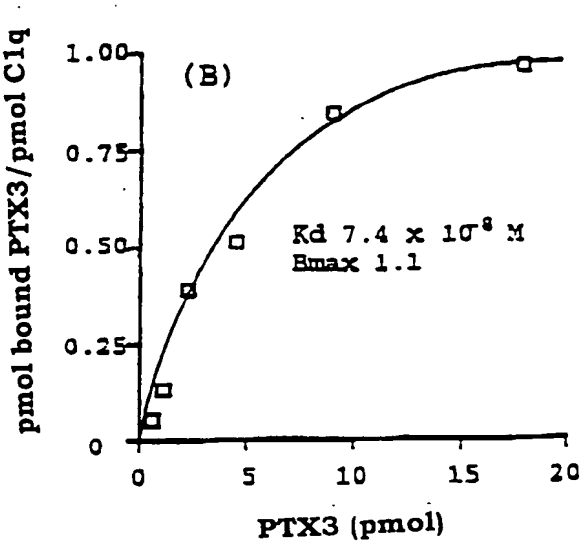

(B)
pmol bound PTX3/pmol C1q
1.00
0.75
0.50
0.25
0
Kd 7.4 x 10⁻⁸ M
Bmax 1.1
0
5
10
15
20
PTX3 (pmol)

FIGURE 3

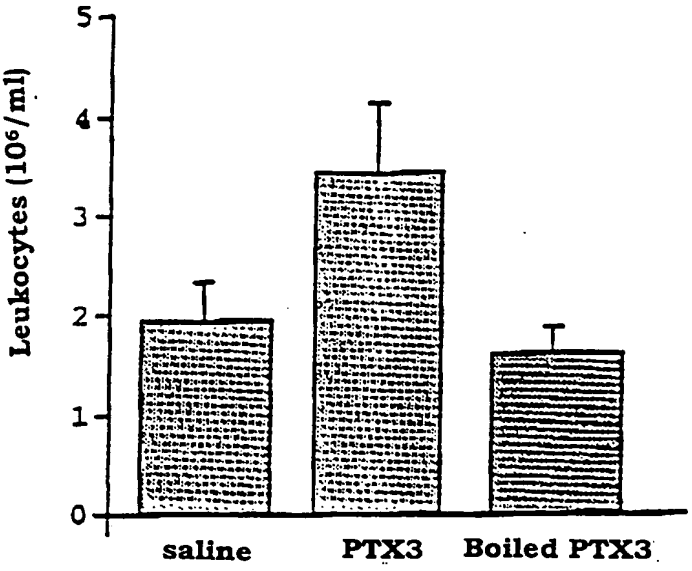
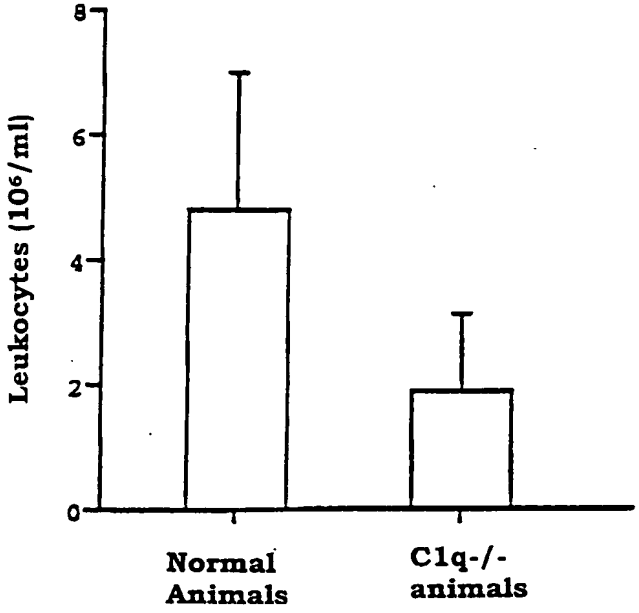


FIGURE 4



Sequence 1

			M	1
HLLAILFCAL	WSAVLAENSD	DYDLMYVNLD	NEIDNGLHPT	41
EDPTPCDCGQ	EHSEWDKLFI	MLENSQMRER	MLLQATDDVL	81
RGELQRLREE	LGRLAESLAR	PCAPGAPAEA	RLTSALDELL	121
QATRDAGRRL	ARMEGAEAQR	PEEAGRALAA	VLEELRQTRA	161
DLHAVQGWAA	RSWLPAGCET	AILFPMRSKK	IFGSVHPVRP	201
MRLESFSACI	WVKATDVLNK	TILFSYGTKR	NPYEIQLYLS	241
YQSIVFVVGG	EENKLVAEAM	VSLGRWTHLC	GTWNSEEGLT	281
SLWVNGELAA	TTVEMATGHI	VPEGGILQIG	QEKNGCCVGG	321
GFDETLAFSG	RLTGFNIWDS	VLSNEEIRET	GGAESCHIRG	361
NIVGWGVTEI	QPHGGAQYVS			381

Sequence 2

```
      ctca aactcagctc acttgagagt ctcctcccgc cagctgtgga aagaactttg      54
cgtctctcca gcaATGCATC TCCTTGCGAT TCTGTTTTGT GCTCTCTGGT CTGCAGTGTT     114
GGCCGAGAAC TCGGATGATT ATGATCTCAT GTATGTGAAT TTGGACAACG AAATAGACAA     174
TGGACTCCAT CCCACTGAGG ACCCCACGCC GTGCGACTGC GGTCAGGAGC ACTCGGAATG     234
GGACAAGCTC TTCATCATGC TGGAGAACTC GCAGATGAGA GAGCGCATGC TGCTGCAAGC     294
CACGGACGAC GTCCTGCGGG GCGAGCTGCA GAGGCTGCGG GAGGAGCTGG GCCGGCTCGC     354
GGAAAGCCTG GCGAGGCCGT GCGCGCCGGG GGCTCCCGCA GAGGCCAGGC TGACCAGTGC     414
TCTGGACGAG CTGCTGCAGG CGACCCGCGA CGCGGGCCGC AGGCTGGCGC GTATGGAGGG     474
CGCGGAGGCG CAGCGCCCAG AGGAGGCGGG GCGCGCCCTG GCCGCGGTGC TAGAGGAGCT     534
GCGGCAGACG CGAGCCGACC TGCACGCGGT GCAGGGCTGG GCTGCCCGGA GCTGGCTGCC     594
GGCAGGTTGT GAAACAGCTA TTTTATTCCC AATGCGTTCC AAGAAGATTT TTGGAAGCGT     654
GCATCCAGTG AGACCAATGA GGCTTGAGTC TTTTAGTGCC TGCATTTGGG TCAAAGCCAC     714
AGATGTATTA AACAAAACCA TCCTGTTTTC CTATGGCACA AAGAGGAATC CATATGAAAT     774
CCAGCTGTAT CTCAGCTACC AATCCATAGT GTTTGTGGTG GGTGGAGAGG AGAACAAACT     834
GGTTGCTGAA GCCATGGTTT CCCTGGGAAG GTGGACCCAC CTGTGCGGCA CCTGGAATTC     894
AGAGGAAGGG CTCACATCCT TGTGGGTAAA TGGTGAACTG GCGGCTACCA CTGTTGAGAT     954
GGCCACAGGT CACATTGTTC CTGAGGGAGG AATCCTGCAG ATTGGCCAAG AAAAGAATGG    1014
CTGCTGTGTG GGTGGTGGCT TTGATGAAAC ATTAGCCTTC TCTGGGAGAC TCACAGGCTT    1074
CAATATCTGG GATAGTGTTC TTAGCAATGA AGAGATAAGA GAGACCGGAG GAGCAGAGTC    1134
TTGTCACATC CGGGGGAATA TTGTTGGGTG GGGAGTCACA GAGATCCAGC CACATGGAGG    1194
AGCTCAGTAT GTTTCAtaaa tgttgtgaaa ctccacttga agccaaagaaa gaaactcac    1254
acttaaaaca catgccagtt gggaaggtct gaaaactcag tgcataatag gaacacttga    1314
gactaatgaa agagagagtt gagaccaatc tttatttgta ctggccaaat actgaataaa    1374
cagttgaagg aaagacattg gaaaaagctt                                    1404
```

SEQUENCE LISTING

<110> sigma tau industrie farmaceutiche riunite s.p.a.

<120> Pharmaceutical compositions containing the long pentraxin PTX3 for the therapy of infectious and inflammatory diseases or tumours, expression vectors containing cDNA coding for PTX3, and use of such v

<130> long human pentraxin

<140>
<141>

<150> RM97A000796
<151> 1997-12-19

<160> 2

<170> PatentIn Ver. 2.0

<210> 1
<211> 381
<212> PRT
<213> HUMAN LONG PENTRAXIN PTX3

<400> 1

```
Met His Leu Leu Ala Ile Leu Phe Cys Ala Leu Trp Ser Ala Val Leu
  1               5                  10                  15

Ala Glu Asn Ser Asp Asp Tyr Asp Leu Met Tyr Val Asn Leu Asp Asn
             20                  25                  30

Glu Ile Asp Asn Gly Leu His Pro Thr Glu Asp Pro Thr Pro Cys Asp
         35                  40                  45

Cys Gly Gln Glu His Ser Glu Trp Asp Lys Leu Phe Ile Met Leu Glu
     50                  55                  60

Asn Ser Gln Met Arg Glu Arg Met Leu Leu Gln Ala Thr Asp Asp Val
 65                  70                  75                  80

Leu Arg Gly Glu Leu Gln Arg Leu Arg Glu Glu Leu Gly Arg Leu Ala
                 85                  90                  95

Glu Ser Leu Ala Arg Pro Cys Ala Pro Gly Ala Pro Ala Glu Ala Arg
            100                 105                 110
```

Leu Thr Ser Ala Leu Asp Glu Leu Leu Gln Ala Thr Arg Asp Ala Gly
115 120 125

Arg Arg Leu Ala Arg Met Glu Gly Ala Glu Ala Gln Arg Pro Glu Glu
130 135 140

Ala Gly Arg Ala Leu Ala Ala Val Leu Glu Glu Leu Arg Gln Thr Arg
145 150 155 160

Ala Asp Leu His Ala Val Gln Gly Trp Ala Ala Arg Ser Trp Leu Pro
165 170 175

Ala Gly Cys Glu Thr Ala Ile Leu Phe Pro Met Arg Ser Lys Lys Ile
180 185 190

Phe Gly Ser Val His Pro Val Arg Pro Met Arg Leu Glu Ser Phe Ser
195 200 205

Ala Cys Ile Trp Val Lys Ala Thr Asp Val Leu Asn Lys Thr Ile Leu
210 215 220

Phe Ser Tyr Gly Thr Lys Arg Asn Pro Tyr Glu Ile Gln Leu Tyr Leu
225 230 235 240

Ser Tyr Gln Ser Ile Val Phe Val Val Gly Gly Glu Glu Asn Lys Leu
245 250 255

Val Ala Glu Ala Met Val Ser Leu Gly Arg Trp Thr His Leu Cys Gly
260 265 270

Thr Trp Asn Ser Glu Glu Gly Leu Thr Ser Leu Trp Val Asn Gly Glu
275 280 285

Leu Ala Ala Thr Thr Val Glu Met Ala Thr Gly His Ile Val Pro Glu
290 295 300

Gly Gly Ile Leu Gln Ile Gly Gln Glu Lys Asn Gly Cys Cys Val Gly
305 310 315 320

Gly Gly Phe Asp Glu Thr Leu Ala Phe Ser Gly Arg Leu Thr Gly Phe
325 330 335

Asn Ile Trp Asp Ser Val Leu Ser Asn Glu Glu Ile Arg Glu Thr Gly
340 345 350

Gly Ala Glu Ser Cys His Ile Arg Gly Asn Ile Val Gly Trp Gly Val
355 360 365

Thr Glu Ile Gln Pro His Gly Gly Ala Gln Tyr Val Ser
370 375 380

<210> 2
<211> 1404
<212> DNA
<213> HUMAN LONG PENTRAXIN PTX3

<400> 2
ctcaaactca gctcacttga gagtctcctc ccgccagctg tggaaagaac tttgcgtctc 60
tccagcaatg catctccttg cgattctgtt ttgtgctctc tggtctgcag tgttggccga 120
gaactcggat gattatgatc tcatgtatgt gaatttggac aacgaaatag acaatggact 180
ccatcccact gaggacccca cgccgtgcga ctgcggtcag gagcactcgg aatgggacaa 240
gctcttcatc atgctggaga actcgcagat gagagagcgc atgctgctgc aagccacgga 300
cgacgtcctg cggggcgagc tgcagaggct gcgggaggag ctgggccggc tcgcggaaag 360
cctggcgagg ccgtgcgcgc cgggggctcc cgcagaggcc aggctgacca gtgctctgga 420
cgagctgctg caggcgaccc gcgacgcggg ccgcaggctg gcgcgtatgg agggcgcgga 480
ggcgcagcgc ccagaggagg cggggcgcgc cctggccgcg gtgctagagg agctgcggca 540
gacgcgagcc gacctgcacg cggtgcaggg ctgggctgcc cggagctggc tgccggcagg 600
ttgtgaaaca gctattttat tcccaatgcg ttccaagaag atttttggaa gcgtgcatcc 660
agtgagacca atgaggcttg agtcttttag tgcctgcatt tgggtcaaag ccacagatgt 720
attaaacaaa accatcctgt tttcctatgg cacaaagagg aatccatatg aaatccagct 780
gtatctcagc taccaatcca tagtgtttgt ggtgggtgga gaggagaaca aactggttgc 840
tgaagccatg gtttccctgg gaaggtggac ccacctgtgc ggcacctgga attcagagga 900
agggctcaca tccttgtggg taaatggtga actggcggct accactgttg agatggccac 960
aggtcacatt gttcctgagg gaggaatcct gcagattggc caagaaaaga atggctgctg 1020
tgtgggtggt ggctttgatg aaacattagc cttctctggg agactcacag gcttcaatat 1080
ctgggatagt gttcttagca atgaagagat aagagagacc ggaggagcag agtcttgtca 1140
catccggggg aatattgttg ggtggggagt cacagagatc cagccacatg gaggagctca 1200
gtatgtttca taaatgttgt gaaactccac ttgaagccaa agaaagaaac tcacacttaa 1260
aacacatgcc agttgggaag gtctgaaaac tcagtgcata ataggaacac ttgagactaa 1320
tgaaagagag agttgagacc aatctttatt tgtactggcc aaatactgaa taaacagttg 1380
aaggaaagac attggaaaaa gctt 1404